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EXAMINER	
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1656	

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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/561,121	DEITERS ET AL.	
	Examiner	Art Unit	
	KAGNEW H. GEBREYESUS	1656	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *24 July 2008*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-45 and 47-61* is/are pending in the application.

4a) Of the above claim(s) *1-39, 44-45, 52-61* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *40-43 and 47-51* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *December 13, 2005* is/are: a)☒ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date *9/5/06 & 11/19/07*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Applicant's election with traverse dated July 24, 2008 in reply to the Office Action dated April 22, 2008 is acknowledged. The **traversal** is on the ground(s) that Applicants do not concede Wang *et al.* is prior art. However Applicants do not provide an explanation for this argument. Furthermore Applicants argue that [3+2] cycloaddition posttranslational modification is not relevant to all of the claims, in contrast to what is stated by the Examiner in the Office Action.

Applicants assert that:

"…these claims share the common inventive concept of the same tRNA synthetase proteins and coding polynucleotides, and further, this inventive concept is a novel special technical feature".

Applicant's reasoning traversing the lack of unity requirement has been carefully considered. However even considering that the tRNA synthetase proteins are the special technical feature, each of the individual aminoacyl-tRNA synthetases or coding polynucleotides sequences is structurally distinct. Furthermore these ORS proteins can have different efficiencies (see definition of preferential aminoacylation ranges between 70-99%) and/or incorporate distinct unnatural amino acids (SEQ ID NO: 48-53 aminoacylate with p-azido-phenylalanine and SEQ ID NO: 54-63 aminoacylate with p-propargyloxyphenylalanine). Thus there is no common technical feature between the various ORS proteins with different structures, different specificities and/or different efficiencies.

Therefore the ORS, OtRNA and the unnatural amino acids can be distinct in their own right. The common technical feature linking the ORS, OtRNA to form a single general inventive concept appears to be a the use of said ORS/O-tRNA to produce proteins comprising an unnatural amino acid which are subsequently post-translationally modified where said modification is a [3+2] cycloaddition between the unnatural amino acid and a second reactive group.

However because the ORS molecules and polynucleotides encoding the same claimed in group X are also encompassed in claims 42-43, 47-51, these claims along with the elected species of SEQ ID NO: 54 encoded by SEQ ID NO: 26 will be examined together. Claims 40-43, 47-51 are present for examination.

Priority

Priority is acknowledged for this application which is a 371 of U.S. National Phase application of International Patent Application NO. PCT/US004/011833 filed on April 16, 2004, which claims priority to and the benefit of US provisional Application 60/479,931 filed June 18, 2003; US provisional Application 60/493,014 filed August 5, 2003; and US provisional Application 60/496,548 filed August 19, 2003. (Please see explanation below).

Information Disclosure Statement

The information disclosure statements filed on September 05, 2006, November 19, 2007 for which a copy of the patent publication have been submitted in this application has been considered.

Oath/Declaration

The oath or declaration submitted on May 23, 2006 has been reviewed and is in compliance with 37 CFR 1.56.

Drawings

The drawings were received on December 13, 2005. These drawings are accepted.

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 40-43, 47-51 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claim 40 (d) recite "at least 50% efficiency as does an O-RS having an amino acid sequence set forth in SEQ ID NO: 45". However the claim does not recite any efficiency for the ORS of SEQ ID NO: 45 or the unnatural amino acid that was used to decipher efficiency. Therefore the claim is indefinite. What is the unnatural amino acid used, the ORS, O-tRNA used to assess efficiency, and what efficiency must be taken as a point of reference in order to compare efficiencies of other ORS molecules?

Furthermore claim 42 part (f) recites a 'conservative variation' of claim 42 part (c) (which is drawn to any polypeptide immunoreactive with an antibody specific to SEQ ID NO: 54). Do these 'conservative variants' also retain immunoreactivity given that conservative variant can have 1, 2, 3…or more changes according to the specification?

Claim 40 is further rejected because the complementary polynucleotides of SEQ ID NO: 20-35 do not encode proteins or ORS molecules (recited in claim 40(a)).

Claim Rejections - 35 USC § 112

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 40, 42, 43, 47-51 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

The specification lacks written description with regards to: the structures of the unnatural amino acid used; the structure of conservative variants of the O-RS molecule of SEQ ID NO: 54 and polynucleotides encoding the same. The ORS molecules that can immunoreact with antibodies specific to the ORS of SEQ ID NO: 54 (claim 42 (c), 47(d)) and conservative variants (47(h); O-RS molecules with 90% identity to any naturally occurring TyrRS from E. coli with at least 2 mutations at selected positions and that retain activity (40 (c)); ORS molecules with aminoacylation efficiency of 50% compared to efficiency of SEQ ID NO: 45 (undisclosed unnatural amino acid or level of efficiency) (40(d); any polynucleotide that hybridizes to SEQ ID NO: 26 or to any polynucleotide that encodes any protein that immunoreacts with the polypeptide of SEQ ID NO: 54 and with a conservative variants thereof (claims 47 (e), (h) – 51).

Factors to be considered in making the determination as to whether one skilled in the art would recognize that the applicant was in possession of the claimed invention as a whole at the time of filing include:

a. Actual reduction to practice;

b. Disclosure of drawings or structural chemical formulas;

c. Sufficient relevant identifying characteristics such as

i. Complete structure,

ii. Partial structure,

iii. Physical and/or chemical properties or

iv. Functional characteristics when coupled with a known or disclosed correlation between function and structure;

d. Method of making the claimed invention;

e. Level of skill and knowledge in the art and

f. Predictability in the art.

While all of these factors are considered, a sufficient number for a *prima facie* case are discussed as they relate to the issues mentioned above.

The specification defines 'a conservative variation' in paragraph [0083]:

"…The term "conservative variant" refers to a translation component, e.g., a conservative variant O-tRNA or a conservative variant O-RS, that functionally performs like the component from which the conservative variant is based, e.g., an O-tRNA or O-RS, but has variations in the sequence. For example, an O-RS will aminoacylate a complementary O-tRNA or a conservative variant O-tRNA with an unnatural amino acid, although the O-tRNA and the conservative variant O-tRNA do not have the same sequence. The conservative variant can have, e.g., one variation, two variations, three variations, four variations, or five or more variations in sequence, as long as the conservative variant is complementary to the corresponding O-tRNA or O-RS."

Therefore, the number of residues to vary is not limited with an upper limit and the specification does not provide a description of any correlation between an ORS structure with unlimited variation and function. For examination purposes any ORS that preferentially aminoacylates a complementary O-tRNA is broadly encompassed in these claims.

The specification teaches a eukaryotic cell comprising the structure of a few ORS molecules and ability to aminoacylate a specific O-tRNA with a specific unnatural amino acid. For example the specification teaches the O-RS of SEQ ID NO: 54 that aminoacylates an O-tRNA with the unnatural amino acid, para-propargyloxyphenylalanine (pPRphe). However the specification does not teach any ORS comprising any conservative variants of the O-RS of SEQ ID NO: 54 or the polynucleotide encoding the O-RS (see claims 40(a), 40(b), 40 (d) and claim 41).

Furthermore the specification does not teach the structure of a conservative variant polypeptide that is immunoreactive with an antibody specific to SEQ ID NO: 54 or a conservative variant which is immunoreactive with an antibody specific to SEQ ID NO: 54 (claim 42 (f), 43, and 47). The skilled artisan cannot predict the structure of a polypeptide variant with unlimited changes to a protein that is immunoreactive with the antibody specific to SEQ ID NO: 54. Claim 47 encompasses polynucleotides of claim 42(c) and variants thereof (claim 47(g),) that encodes any of the polypeptides including the immunoreactive polypeptides recited in claim 42(c). However the specification does not permit one of skill in the art to predict the structure of all polynucleotide sequences that encode polypeptides that are conservative variants to a protein that can immunoreact with the antibody produced for the ORS of SEQ ID NO: 54 and variants with unlimited changes as recited above (47(h), 48-51).

Moreover, claim 40 is drawn to a eukaryotic cell comprising an ORS molecule of SEQ ID NO: 54 and conservative variant or ORS variants with at least 90% identity to any naturally occurring E. coli Tyr-RS structure with at least two changes at positions corresponding to position Tyr37, Asn126, Asp182, Phe183 and Leu186 in *E. coli* tyrosyl tRNA synthetase that preferentially aminoacylate any O-tRNA with **any unnatural amino acid** (40 (c)). The specification teaches a few ORS molecules such as the ORS of SEQ ID NO: 54 that aminoacylates the O-tRNA of SEQ ID NO: 65 with para-propargyloxyphenylalanine (pPRphe) or the ORS of SEQ ID NO: 86 that aminoacylates a corresponding O-tRNA with p-acetylphenylalanine. However the claims encompass cells comprising 'conservative variant' of SEQ ID NO: 54 or a variants of any *E. coli* Tyr-RS with at least 90% identity and where said ORS can aminoacylate an O-tRNA with any structurally undefined unnatural amino acid.

However at the time the instant invention was filed Applicants were not in possession of the above broadly claimed Tyr-ORS variants. Moreover, the art teaches that O-RS molecules with specific structures can only accommodate specific unnatural amino acid(s), not any unnatural amino acid in their active site. The specification and the art teach that determination of ORS structures that aminoacylate a corresponding O-tRNA with a specific unnatural amino acid must be empirically determined and cannot be predicted *a priori*.

Furthermore the structure of the O-RS is dictated by the structure of the unnatural amino acid(s) to be preferentially aminoacylated onto a corresponding O-tRNA.

Claim 40 (d) is drawn to eukaryotic cells comprising any ORS that preferentially aminoacylates any O-tRNA with any unnatural amino acid with an efficiency of at least 50% relative to the efficiency of the ORS of SEQ ID NO: 45 (no specific unnatural amino acid

mentioned therefore the claim encompasses any unnatural amino acid). However no structure or functional characteristics coupled with a known or disclosed correlation between function and structure can be made for any member of the genus of polypeptides with the limitation of 'at least 50% efficiency' relative to the structure of SEQ ID NO: 54 (elected species). Preferential aminoacylation efficiency must be determined empirically for each member of the genus in the presence of a known O-RS, O-tRNA and a known unnatural amino acid. One of skill in the art cannot predict the structure of an ORS polypeptide that functions with at least 50% efficiency relative to an efficiency that varies depending on the unnatural amino acid, the O-tRNA even with a known O-RS such as SEQ ID NO: 54.

Furthermore claim 42 (c) and 42 (f) are drawn to O-RS polypeptides that are specifically immunoreactive with an antibody specific for a polypeptide of SEQ ID NO: 54 and conservative variants.

The specification teaches the structure of the O-RS of SEQ ID NO: 54 and how these O-RS molecule can be used as immunogens to produce specific antibodies. However the specification does not teach the structure of an epitope that is common to said ORS variants encompassed in the claimed genus of proteins (in claim 42(c)) or in the variants claimed in claim 42(h) with any identifying characteristic such as a complete structure, partial structure, physical or chemical property to lead one skilled in the art to conclude that Applicants were in possession of the claimed invention.

Furthermore, the skilled artisan cannot predict the structure of the genus of proteins based on the disclosure of the ORS of SEQ ID NO: 54 because any native or chimeric protein which

comprises an epitope structure identical to at least one epitope found in SEQ ID NO: 54 can be broadly encompassed in the claims.

Given this lack of description of representative species encompassed by the claimed genus, the specification fails to sufficiently describe the invention in such full, clear, concise, and exact terms that a skilled artisan would recognize that applicants were in possession of the claimed invention.

Claims 40-43, 47-51 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for eukaryotic cells comprising O-RS molecules comprising SEQ ID NO: 54 and O-RS molecules encoded by SEQ ID NO: 26 that aminoacylates a corresponding O-tRNA (SEQ ID NO: 65) with para-propargyloxyphenylalanine (pPR-1), does not reasonably provide enablement for eukaryotic cells comprising any ORS molecule including SEQ ID NO: 54, conservative variants and any ORS that is at least 90% identical to any E. coli Tyr-tRNA synthetase (TyrRS) wherein said O-RS molecules aminoacylate any O-tRNA with any unnatural amino acid.

The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

Factors to be considered in determining whether undue experimentation is required, are summarized ln re Wands (858 F.2d 731, 8 USPQ 2nd 1400 (Fed. Cir. 1988). The Wands factors are: (a) the nature of the invention, (b) the relative skill of those in the art, (c) the state of the prior art, (d) the predictability or unpredictability of the art, (e) the amount of direction or

guidance presented, (f) the presence or absence of working example, (g) the breadth of the claim, (h) the quantity of experimentation necessary.

The nature of the invention is directed to eukaryotic cells comprising specific ORS molecules (e.g. SEQ ID NO: 48-63) and corresponding amber suppressor $tRNA_{CUA}$ (O-tRNA of SEQ ID NO: 65) wherein said ORS molecules preferentially aminoacylate said O-tRNA with para-propargyloxyphenylalanine (pPRO-Phe). Said O-tRNA charged with pPRO-Phe incorporates pPRO-Phe into a protein.

The state of the prior art teaches that preferential aminoacylation with any specific unnatural amino acid requires the use of specific ORS molecules (or group of ORS molecules with common structural characteristics) in concert with a corresponding amber suppressor O-tRNA molecule which incorporates the unnatural amino acids into a protein.

However the claims are drawn to eukaryotic cells comprising an ORS molecule of SEQ ID NO: 54, 'conservative variants' or a protein with at least 90% identity to any naturally occurring Tyr-RS (40 (c)) that comprises at least two changes at positions corresponding to position Tyr37, Asn126, Asp182, Phe183 and Leu186 in *E. coli* tyrosyl tRNA synthetase, that preferentially aminoacylate any O-tRNA (claim 40) or the O-tRNA of SEQ ID NO: 65 (claim 41) with <u>any unnatural amino acid</u>.

The specification, supported by the prior art teaches that aminoacylation specificity of orthogonal tRNA synthetases (O-RS) to specific unnatural amino acids results from structural modification introduced in the amino acid binding pocket and/or other regions of a native tRNA synthetase. Such ORS will differ from their native counterparts by acquiring increased specificity for an unnatural amino acid compared to specificity for their natural amino acid

substrates. However neither the prior art nor the instant specification teach how to make or use eukaryotic cells comprising any specific O-RS including the O-RS of SEQ ID NO: 54, that can acquire the ability to aminoacylate any possible type of unnatural amino acids onto an O-tRNA.

The art teaches that mutations at positions other than the amino acid binding pocket of an ORS can bring about changes in amino acid specificity. For example US 5,370,995 (Hennecke et al) teaches that a mutation at position 294 of *E. coli* phenylalanine tRNA synthetase results in altered specificity. Hennecke et al teach that an alanine to glycine substitution at position 294 (Gly294) results in incorporation of para-fluoro-phenylalanine instead of phenylalanine into a protein.

Moreover the claims encompass cells comprising O-RS molecules with any structure that can aminoacylate a corresponding O-tRNA with any unnatural amino acid with at least 50% efficiency compared to the efficiency of the ORS of SEQ ID NO: 45 (claim 40 (d)). However while the specification only provides guidance and examples of making O-RS molecules (SEQ ID NO: 54) that can aminoacylate an O-tRNA with pPR. The specification does not provide guidance on how to make and use ORS molecules that can aminoacylate any O-tRNA with any unnatural amino acid with any efficiency.

Furthermore claim 42 (c), 42 (f), 43, 47-51 encompass O-RS polypeptides or polynucleotides encoding the same with at least 90% identity to any naturally occurring *E. coli* TyrRS and 'conservative variants' or polypeptides with at least 20 contiguous amino acids and 'conservative variants' (claim 42(f)). However the scope of 'conservative variation' is not limited to any degree of variation of the ORS sequence, therefore it encompasses any sequence with any structure which performs as the ORS from which it is derived. However applicants have

not taught how to make and how to use the scope of all possible ORS variants or polynucleotides encoding the same. For example one of skill will require undue amount experimentation to identify the unnatural amino acid that can be accommodated by a particular variant of an ORS.

Furthermore claims 42(f), 43, 47(h)-51 also encompass any polypeptide or polynucleotide encoding the same that are specifically immunoreactive with an antibody specific for a polypeptide of SEQ ID NO: 54 (claim 47(h)-51) and conservative variants of said molecules. However while certain truncated variants of SEQ ID NO: 54 such as fragments containing the same epitopes found in SEQ ID NO: 54 that immunoreact with an antibody for SEQ ID NO: 54, the specification does not teach how to make and use all possible polypeptide variants derived from SEQ ID NO: 54 which would immunoreacts with an antibody specific for SEQ ID NO: 54. (thus functionally performs like the component from which the conservative is based upon as defined in the specification).

Without sufficient guidance, determination of eukaryotic cells comprising ORS molecules of SEQ ID NO: 54 or molecules with 90% identity to any E. coli Tyr-ORS and conservative variants thereof that can aminoacylate an O-tRNA with any unnatural amino acid (claims 40, 41) or any polypeptide or polynucleotide encoding the same that immunoreacts with an antibody for SEQ ID NO: 54 (claim 42(f), 43, 47(d) 47(h) is unpredictable. Furthermore the specification does not provide guidance with regards to polynucleotides or conservative variants that encode O-RS molecules with 90% identity to an *E. coli* TyrRS and can 'preferentially' aminoacylate an OtRNA with any type of unnatural amino acid. Furthermore the specification does not teach how to make an ORS with at least 50% aminoacylation efficiency compared to the efficiency of SEQ ID NO: 45 to aminoacylate any unnatural amino acid. The efficiency

obtained using a specific ORS/O-tRNA pair will vary depending on the unnatural amino used. However the claims (claim 40(d), 41) encompass eukaryotic cells comprising any ORS that aminoacylates an O-tRNA with at least 50% aminoacylation efficiency with any unnatural amino acid. To accomplish the skilled artisan would have to first determine aminoacylation efficiency for a composition comprising any ORS/OtRNA pair and any possible unnatural amino acid and then identify all possible ORSs that aminoacylate with 50% efficiency to each ORS/OtRNA, unnatural amino acid composition.

Without sufficient guidance with regards to the specific structure of the ORS/OtRNA pair and the unnatural amino acid, determination of ORS with 50% efficiency is unpredictable and the experimentation left to those skilled in the art is unnecessarily, and improperly, extensive and undue. See In re Wands 858 F.2d 731, 8 USPQ2nd 1400 (Fed. Cir, 1988).

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (a) the invention was known or used by others in this country, or patented or described in a printed publication in this or a foreign country, before the invention thereof by the applicant for a patent.

Claims 40-43, 47-51 are rejected under 35 U.S.C. 102 (a) as being anticipated by Sakamoto et al.. (2002)

Sakamoto et al teach CHO-Y cells (Chinese hamster ovary cells) or human embryonic kidney 293 cells transfected with a vector expressing tyrosyl-ORS and a corresponding O-tRNA where said Tyr-ORS preferentially aminoacylates said O-tRNA with an unnatural amino acid

analogue of L-tyrosine (3-iodo-L-tyrosine) into a protein. Figs. 4a and 4b show production of the Ras protein 4(a) and EGFR 4(b) comprising an unnatural amino acid. In the presence of OtRNA from *Bacillus stearothermophilus* and *E. coli* Tyr-O-RS a significant amount of EGFR protein was produced from an EGFR gene comprising an amber suppressor codon compared to from a wild type EGFR gene. Furthermore Fig. 7 shows CHO-Y cells expressing a Ras protein comprising the unnatural amino acid from a Ras gene comprising an amber suppressor codon wherein said cells comprise a *Bacillus stearothermophilus* suppressor tRNA (O-tRNA) and *E. coli* Tyr-O-RS. A significant amount of Ras protein is produced only in the presence of the unnatural amino acid, 3-iodo-L-tyrosine (see lane 1) while no Ras protein was observed in the absence of 3-iodo-L-tyrosine indicating preferential aminoacylation of the *Bacillus stearothermophilus* suppressor tRNA by the *E. coli* Tyr-O-RS. Given that the definition of 'conservative variant' broadly encompasses any ORS the disclosure of Sakamoto et al anticipates claims 40-43, 47-51 (specifically what is recited in claims 40(b), 40(d), 41, 42(c), 42(f), 43, 47-51). Claims 40, 41, 43(f), 47(h)-51 encompass conservative variants of the O-tRNA of SEQ ID NO: 65 from *E. coli,* 'conservative variants' of the ORS of SEQ ID NO: 54 or the polynucleotide encoding the same, vectors and eukaryotic host cells comprising said ORS/O-tRNA. Thus Sakamoto et al's CHO-Y cells (Chinese hamster ovary cells) or human embryonic kidney 293 cells transfected with vectors expressing tyrosyl-ORS and a corresponding O-tRNA where said Tyr-ORS preferentially aminoacylates said O-tRNA with the unnatural amino acid analogue of L-tyrosine (3-iodo-L-tyrosine) into a protein anticipate the instant claims.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The factual inquiries set forth in *Graham* **v.** *John Deere Co.*, 383 U.S. 1, 148 USPQ 459 (1966), that are applied for establishing a background for determining obviousness under 35 U.S.C. 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.
2. Ascertaining the differences between the prior art and the claims at issue.
3. Resolving the level of ordinary skill in the pertinent art.
4. Considering objective evidence present in the application indicating obviousness or nonobviousness.

Claims 40-43, 47-51 as it pertains to elements 40(b), 40(d), 41, 42(c), 42(f), 43 and 47-51 are rejected under 35 U.S.C. 103(a) as being unpatentable over Kiga et al. (2002, in IDS) in view of Dougherty et al (Unnatural amino acids as probes of protein structure and function. Current opinions in Chemical Biology 2000, 4:645-652) further in view of Sakamoto et al. (Site specific Incorporation of an unnatural amino acid into proteins in mammalian cells Nucleic Acid Research, 2002, Vol. 30 No. 21).

Kiga et al teach a eukaryotic translation system (wheat germ cell free translation system) in which a variant of *E. coli* tyrosyl tRNA synthetase and a suppressor tRNA from *E. coli* were used to incorporate 3-iodo-L-tyrosine (an unnatural amino acid analogue of L-tyrosine) into a protein. As defined in the specification a 'conservative variant' of an ORS can comprise 1, 2,

3… or more variations thus can encompass any type of ORS that preferentially aminoacylates any complementary O-tRNA. While Kiga et al's teach an ORS variant wherein said ORS preferentially aminoacylates a corresponding O-tRNA with 3-iodo-L-tyrosine, they do not teach a eukaryotic cell comprising an ORS variant wherein said ORS preferentially aminoacylates a corresponding O-tRNA in a eukaryotic cell *per se*.

Sakamoto et al teach CHO-Y cells (Chinese hamster ovary cells) or human embryonic kidney 293 cells transfected with a vector expressing tyrosyl-ORS from *E. coli* and a corresponding O-tRNA from *Bacillus stearothermophilus* where said Tyr-ORS preferentially aminoacylates said O-tRNA with an unnatural amino acid, 3-iodo-L-tyrosine into a protein (discussed above).

Therefore providing a yeast eukaryotic cell comprising the ORS and O-tRNA of Kiga et al ***would have been obvious*** in view of Sakamoto et al's disclosure discussed above.

One of skill in the art would have ***a reasonable expectation of success*** because at least two different eukaryotic cells CHO-Y and human embryonic kidney 293 cells comprising an ORS and O-tRNA were taught by Sakamoto et al. Furthermore one of ordinary skill in the art ***would be_motivated*** to provide a eukaryotic cell comprising an ORS that preferentially aminoacylates an O-tRNA because proteins comprising unnatural amino acids can be made for various applications including those described in Dougerty et al. (see below).

Dougherty et al teach injection of a suppressor tRNA aminoacylated with an unnatural amino acid and a polynucleotide encoding a protein of interest in *Xenopus* oocytes (a eukaryotic cell) (see fig.1). Said unnatural amino acid is incorporated in a specific position in the expressed protein. (Shown in fig. 2 page 648). Furthermore they teach that incorporation of biophysical

probes such as fluorescent unnatural amino acids in the neurokinin-2 receptor can be valuable in studying distance using fluorescence energy transfer information relating the location of the agonist-binding site to selected residues in a receptor (see page 649, first column). Thus, Dougherty et al's study provides at least one motivation for producing eukaryotic cell comprising an ORS that preferentially aminoacylates an O-tRNA.

Claims 40-43, 47-51 are rejected under 35 U.S.C. 103(a) as being unpatentable over US PAT 7083970 (Schultz et al), (in IDS) further in view of Kiga et al. (in IDS) further in view of Dougherty et al..

The instant application recites eukaryotic cells comprising 'conservative variants' of SEQ ID NO: 54 or 'conservative variants' of the polynucleotide of SEQ ID NO: 26 wherein the encoded ORS can preferentially aminoacylate a corresponding O-tRNA.

Schultz et al teach a cell comprising an ORS that preferentially aminoacylates an O-tRNA with an unnatural amino acid wherein site-selective incorporation of one or more unnatural amino acids such as 3-substituted tyrosine and 4-iodo-substituted tyrosine into proteins in vivo (fig. 31). Schultz et al teach the use of specific vectors to introduce the nucleic acid target that comprises generic expression cassettes containing at least one independent terminator sequence, sequences permitting replication of the cassette ***in eukaryotes cells, or prokaryotes cells, or both, (e.g., shuttle vectors)*** and selection markers for both prokaryotic and ***eukaryotic systems*** (see column 48 line 60-67 and column 49 line 1-27 and line 43-67 describing vectors comprising the ORS/OtRNA used for ***transfecting*** cells. Furthermore transgenic organisms comprising said cells are also envisioned in column 49 lines 14-15 of Schultz et al.

However Schultz et al do not exemplify a eukaryotic system comprising an ORS and OtRNA that preferentially aminoacylate the corresponding O-tRNA with an unnatural amino acid.

However Kiga et al teach a eukaryotic translation system (wheat germ cell free translation system) in which a variant of *E. coli* tyrosyl tRNA synthetase that preferentially aminoacylates a corresponding suppressor tRNA from *E. coli* were used to incorporate 3-iodo-L-tyrosine (an unnatural amino acid analogue of L-tyrosine) into a protein. While Kiga et al's teach a variant of the ORS SEQ ID NO: 54 wherein said ORS preferentially aminoacylates a corresponding O-tRNA they do not teach a eukaryotic cell comprising the same *per se*.

Therefore it would have been ***obvious*** to produce cells comprising an ORS that preferentially aminoacylates an O-tRNA using the combination of teaching by Kiga et al and the eukaryotic expression vectors described in Schultz et al.

One of ordinary skill in the art ***would have a reasonable expectation of success*** in producing eukaryotic cells comprising an ORS that preferentially aminoacylates an O-tRNA because Kiga et al's teaches that a eukaryotic system is capable to supporting preferential aminoacylation of an O-tRNA by an ORS. Furthermore Schultz et al provide vectors comprising polynucleotides encoding the ORS and O-tRNA necessary to transfect eukaryotic cells thus produce eukaryotic host cells comprising the ORS and O-tRNA. One of ordinary skill in the art ***would be motivated*** to produce such a cell because of the various useful applications described by Dougherty et al. discussed above.

Conclusion

No claim is allowed.

Relevant publication:

Edwards et al. A Bacterial Amber Suppressor in Saccharomyces cerevisiae Is selectively Recognized by a Bacterial Aminoacyl-tRNA Synthetase. Molecular and cell Biology. April 1990, pages 1633-1641). Edward et al teach that E. coli ORS can aminoacylate and E. coli OtRNA in Saccharomyces cerevisiae. However they do not teach aminoacylation with an unnatural amino acid.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to KAGNEW H. GEBREYESUS whose telephone number is (571)272-2937. The examiner can normally be reached on 8:30am-5:30pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Jon Weber can be reached on 571-272-0925. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300. Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

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